

M2-IM were able to induce fibroblast activation *in vitro* mediated by an enhanced TGF- $\beta$ 1 expression suggesting a profibrotic role of M2-IM. Specific depletion of hybrid AM using intranasal administration of clodrosome increased radiation-induced fibrosis score and enhanced M2-IM infiltration suggesting a protective role of hybrid AM.

**Conclusion:** These present study shows a dual and opposite contribution of alveolar *versus* interstitial macrophages in radiation-induced fibrosis and identify M2-IM as a potential therapeutic target to treat radiation-induced fibrosis.

#### EP-2045

**In vivo monitoring of skin collagen state by multiphoton microscopy in the course of irradiation**

N.D. Gladkova<sup>1</sup>, V.V. Dudenkova<sup>1,2</sup>, V.V. Elagin<sup>1</sup>, K.V. Babak<sup>3</sup>, A.V. Maslennikova<sup>3,4</sup>

<sup>1</sup>State Medical Academy, Research institute of biomedical technologies, Nizhny Novgorod, Russian Federation

<sup>2</sup>Lobachevsky State University, General physics, Nizhny Novgorod, Russian Federation

<sup>3</sup>Lobachevsky State University, Biophysics, Nizhny Novgorod, Russian Federation

<sup>4</sup>State Medical Academy, Oncology and Radiotherapy beam diagnostics, Nizhny Novgorod, Russian Federation

**Purpose or Objective:** Adverse events in normal tissues during and after a course of cancer radiation treatment are one of the most pressing problems of modern radiation oncology. From among numerous works in this field, there are but a few concerned with the radiation-induced alterations of collagen, the processes of its degradation and subsequent remodeling. A new imaging technique - multiphoton microscopy (MPM) allows studying tissue collagen state on fibers and bundles level without additional staining due to second harmonic generation (SHG) phenomenon. The method has the key advantage of a potential in vivo application. This study's objective was in vivo evaluation of changes occurring at rat's skin collagen upon the exposure of conventional irradiation.

**Material and Methods:** Rat's ear was chosen as a model for detecting collagen changes. Experiments were carried out under Nizhny Novgorod Medical Academy ethical committee permission. Three male animals, 2 months old at the time of experiment, were used. Rat's ear was irradiated under general anesthesia (Zoletyl, 50 mg/kg, Virbac Sante Animale, France) by a Co60 unit Terabalt (UJP, Czech Republic) by a local field with single dose of 2 Gy up to the total dose of 24 Gy. The 3D imaging of collagen structure was performed by MPTflex (JenaLab, Germany) - a system for in vivo optical biopsies based on near infrared femtosecond laser technology. MPM imaging was carried out two times a week beginning from the first day of irradiation and once a week for three months after its completion. Cross-sectional images were obtained beginning from the horny layer with the step of 5  $\mu$ m up to the total depth of 100  $\mu$ m. Excitation was implemented with a pulsed (200 fs) titanium-sapphire laser at a wavelength of 740 nm and a pulse repetition frequency of 80 MHz; SHG collagen imaging was performed at 373-387 nm. Cross-sectional images of 512x512 pixels were obtained; the field size was 130x130  $\mu$ m. Numerical processing of the images was performed by ImageJ program. Mean fluorescence intensity and its standard deviation was calculated for all images. Coefficient S (a ratio of standard deviation/mean fluorescence intensity) was used for evaluation of collagen state.

#### Results:

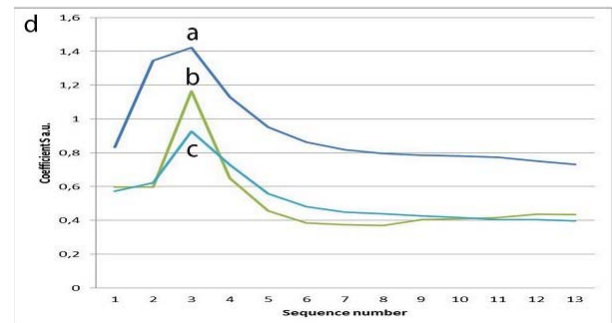
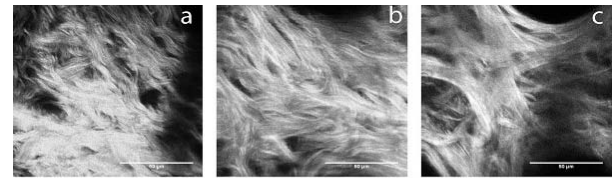


Fig. 1. Examples of the MPM images of rat's ear: a - before irradiation, b - 24 Gy, c - 24 Gy, 3 month after irradiation, d - correspondent coefficient S.

Visual evaluation of MPM images demonstrated no noticeable changes of collagen packing and structure independent on the dose and time from radiation beginning (Fig.1, a, b, c). Numerical processing revealed subtle, but clear differences of coefficient S between intact and irradiated collagen. After radiation beginning, a decrease of magnitude of coefficient S and the decrease of tilt angle of the graph was observed (Fig.1 d). In a month after radiation completion, a magnitude remained decreased, but tilt angle of the graph returned to the initial level (Fig.1 d).

**Conclusion:** Numerical processing of MPM-images demonstrated changes of optical properties of collagen upon expose of clinically relevant doses of gamma-irradiation. The radiobiological interpretation of these changes require further study.

#### EP-2046

**Modulation of radiation-induced oral mucositis (mouse) by dermatan sulfate**

S. Gruber<sup>1</sup>, E. Bozsaky<sup>1</sup>, K. Frings<sup>1</sup>, M. Arnold<sup>1</sup>, V. Gernedl<sup>1</sup>, S. Hetzendorfer<sup>1</sup>, J. Mayer<sup>1</sup>, S. Morava<sup>1</sup>, S. Pfaffinger<sup>1</sup>, P. Kuess<sup>2</sup>, W. Dörr<sup>1</sup>

<sup>1</sup>Medical University of Vienna, Department of Radiotherapy-ATRAB - Applied and Translational Radiobiology and Christian Doppler Laboratory for Medical Radiation Research for Radiation Oncology, Vienna, Austria

<sup>2</sup>Medical University of Vienna, Department of Radiation Oncology and Christian Doppler Laboratory for Medical Radiation Research for Radiation Oncology, Vienna, Austria

**Purpose or Objective:** Oral mucositis is the most frequently occurring, dose limiting early adverse event of head-and-neck cancer radio(chemo)therapy. The purpose of the present study was to quantify the mucoprotective effect of dermatan sulfate (DS), and to characterise the associated changes in the expression of markers for epithelial proliferation, cell junctions, inflammation and hypoxia.

**Material and Methods:** The study comprises a functional and a histological arm. For the functional investigations, mice were irradiated with 5x3 Gy/week over one (days 0-4) or two weeks (days 0-4, 7-11). Each protocol was concluded by irradiation with graded top-up doses (day 7/14), to generate complete dose-effect curves. Daily doses of DS (4 mg/kg subcutaneously) were applied over varying time intervals. Mucosal ulceration, was analysed as clinically relevant endpoint during the functional studies. In the histological study, groups of three mice were sacrificed every second day, the tongues were excised and subjected to histological/immunohistochemical processing.

**Results:** DS significantly increased isoeffective doses for the induction of oral mucositis in almost all protocols, and